

=> s pHL1 vector
L1 1 PHL1 VECTOR

=> d l1 ibib ab

L1 ANSWER 1 OF 1 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1988:125827 HCAPLUS
DOCUMENT NUMBER: 108:125827
TITLE: Gene transfer system for the phytopathogenic fungus
Ustilago maydis
AUTHOR(S): Wang, Jun; Holden, David W.; Leong, Sally A.
CORPORATE SOURCE: Dep. Plant Pathol., Univ. Wisconsin, Madison, WI,
53706, USA
SOURCE: Proceedings of the National Academy of Sciences of the
United States of America (1988), 85(3), 865-9
CODEN: PNASA6; ISSN: 0027-8424
DOCUMENT TYPE: Journal
LANGUAGE: English

AB A selectable marker for transformation was constructed by transcriptional fusion of a U. maydis heat shock gene promoter with the hygromycin B phosphotransferase gene of Escherichia coli. U. maydis was transformed to hygromycin B resistance by polyethylene glycol-induced fusion of spheroplasts following exposure to plasmid DNA that carried the marker gene. Transformation frequencies of 50 and 1000 transformants per .mu.g of DNA per 2 .times. 10⁷ spheroplasts were obtained for circular and linear vector DNA, resp. In the majority of transformants, the vector was integrated at a single chromosomal site, in either single copy or tandem duplication, as detd. by Southern hybridization anal. of electrophoretically sepd. chromosomes and of restriction-endonuclease-cleaved DNA. The predominant form (82%) of vector integration was by nonhomologous recombination; the remainder carried the plasmid at the homologous heat shock gene locus. No evidence for gene conversion or gene replacement was obtained in 28 transformants. Hygromycin B phosphotransferase activity and resistance to hygromycin B were roughly correlated with the copy no. of the integrated vector at the homologous location. Transforming DNA was stably maintained during mitosis and meiosis. This transformation procedure and assocd. vector should permit the cloning of genes by direct complementation in U. maydis.

=> FIL STNGUIDE
COST IN U.S. DOLLARS

SINCE FILE	TOTAL
ENTRY	SESSION
8.76	9.24

FULL ESTIMATED COST

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)

SINCE FILE	TOTAL
ENTRY	SESSION
-0.69	-0.69

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FILE CONTAINS CURRENT INFORMATION.
LAST RELOADED: Apr 23, 2004 (20040423/UP).

=> d l1 ibib ab

YOU HAVE REQUESTED DATA FROM FILE 'HCAPLUS' - CONTINUE? (Y)/N:y

L1 ANSWER 1 OF 1 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1988:125827 HCAPLUS
DOCUMENT NUMBER: 108:125827

TITLE: Gene transfer system for the phytopathogenic fungus
 Ustilago maydis
 AUTHOR(S): Wang, Jun; Holden, David W.; Leong, Sally A.
 CORPORATE SOURCE: Dep. Plant Pathol., Univ. Wisconsin, Madison, WI,
 53706, USA
 SOURCE: Proceedings of the National Academy of Sciences of the
 United States of America (1988), 85(3), 865-9
 CODEN: PNASA6; ISSN: 0027-8424
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB A selectable marker for transformation was constructed by transcriptional fusion of a U. maydis heat shock gene promoter with the hygromycin B phosphotransferase gene of Escherichia coli. U. maydis was transformed to hygromycin B resistance by polyethylene glycol-induced fusion of spheroplasts following exposure to plasmid DNA that carried the marker gene. Transformation frequencies of 50 and 1000 transformants per .mu.g of DNA per 2 .times. 10⁷ spheroplasts were obtained for circular and linear vector DNA, resp. In the majority of transformants, the vector was integrated at a single chromosomal site, in either single copy or tandem duplication, as detd. by Southern hybridization anal. of electrophoretically sepd. chromosomes and of restriction-endonuclease-cleaved DNA. The predominant form (82%) of vector integration was by nonhomologous recombination; the remainder carried the plasmid at the homologous heat shock gene locus. No evidence for gene conversion or gene replacement was obtained in 28 transformants. Hygromycin B phosphotransferase activity and resistance to hygromycin B were roughly correlated with the copy no. of the integrated vector at the homologous location. Transforming DNA was stably maintained during mitosis and meiosis. This transformation procedure and assocd. vector should permit the cloning of genes by direct complementation in U. maydis.

=> file hcaplus medline biosis embase biotechds
 COST IN U.S. DOLLARS

SINCE FILE	TOTAL
ENTRY	SESSION
0.06	14.32

FULL ESTIMATED COST

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)

SINCE FILE	TOTAL
ENTRY	SESSION
0.00	-1.38

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FILE 'MEDLINE' ENTERED AT 10:53:35 ON 29 APR 2004

FILE 'BIOSIS' ENTERED AT 10:53:35 ON 29 APR 2004
 COPYRIGHT (C) 2004 BIOLOGICAL ABSTRACTS INC.(R)

FILE 'EMBASE' ENTERED AT 10:53:35 ON 29 APR 2004
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FILE 'BIOTECHDS' ENTERED AT 10:53:35 ON 29 APR 2004
 COPYRIGHT (C) 2004 THOMSON DERWENT AND INSTITUTE FOR SCIENTIFIC INFORMATION

=> s pHLB29
 L2 1 PHLB29

=> d l2 ibib ab

L2 ANSWER 1 OF 1 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
 ACCESSION NUMBER: 2004-05981 BIOTECHDS
 TITLE: Alkaline lipase useful as laundry detergent, isolated from

Vibrio metschnikovii RH530 N-4-8;
recombinant enzyme production in Escherichia coli

AUTHOR: JIN G; JHON S; LEE H; RHO H
PATENT ASSIGNEE: CJ CORP
PATENT INFO: WO 2004001029 31 Dec 2003
APPLICATION INFO: WO 2003-KR1227 23 Jun 2003
PRIORITY INFO: KR 2002-35410 24 Jun 2002; KR 2002-35410 24 Jun 2002
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2004-082499 [08]

AB DERWENT ABSTRACT:

NOVELTY - An alkaline lipase (I) isolated from Vibrio metschnikovii RH530 N-4-8 comprising a fully defined sequence of 185 amino acids (S1) as given in the specification, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (1) polynucleotide (II) encoding (S1); (2) recombinant vector (III) comprising (II); (3) host cell (IV) transformed by (III); and (4) detergent comprising (I).

BIOTECHNOLOGY - Preparation: (I) is produced by culturing (IV) (claimed). Preferred Polynucleotide: (II) comprises a fully defined sequence of 555, 798 or 2578 base pairs as given in the specification. Preferred Recombinant Vector: (III) is pHL1, pHLB29 or pHAAH38.

USE - (I) is useful as an enzyme for laundry detergent (claimed).

ADVANTAGE - (I) has high residual enzyme activity and high compatibility.

EXAMPLE - Culture medium comprising tryptone, yeast extract, sodium chloride in sodium carbonate buffer was used for culturing Vibrio metschnikovii RH530 N-4-8, at 30 degreesC. The cells were collected and treated with lysozyme to lyse the cell. The resultant product was treated with phenol and chloroform to remove protein, and a precipitate was removed by centrifugation. A Vibrio chromosomal DNA was obtained from the supernatant. The obtained chromosomal DNA was cut with a restriction enzyme HindIII to be recombined with a cloning vector pUC19, followed by transforming Escherichia coli HB101, thus cloning a DNA fragment containing a 3.2 kb alkaline lipase gene. The resulting recombinant vector was referred to as a vector pHL1. After treatment with the restriction enzyme HindIII, an electrophoresis with 1% agarose gel was performed. The agarose gel electrophoresis showed that the alkaline lipase gene was cloned. To confirm that a DNA fragment containing an alkaline lipase gene derived from V. metschnikovii, which is contained in a recombinant vector pHL1, is identical with the gene from V. metschnikovii, Southern blotting was performed. DNA of 3.2 kb was treated with an exonuclease Bal31 to subclone the same in a minimum length required for expression of a lipase. Production of the lipase was confirmed by formation of a clear halo, and the result of subcloning showed that 2.6 kb DNA fragment was necessary for lipase activity. The recombinant vector containing such a gene having a minimum length was referred to as pHLB29. DNA of 2.6 kb fragment was subcloned in a direction opposite to that of a SmaI site of pUC19, and referred to as pHAAH38. Although the 2.6 kb DNA fragment was subcloned in a reverse direction relative to a lac promoter, pHAAH38 produced a clear halo at a tricaprillin culture medium, confirming that an alkaline lipase promoter existed in the 2.6 kb DNA fragment and the promoter used when it is transcribed from E. coli. (35 pages)

=> d his

(FILE 'HOME' ENTERED AT 10:50:33 ON 29 APR 2004)

FILE 'STNGUIDE' ENTERED AT 10:50:38 ON 29 APR 2004

FILE 'HOME' ENTERED AT 10:50:41 ON 29 APR 2004

FILE 'HCAPLUS, MEDLINE, BIOSIS, EMBASE, BIOTECHDS' ENTERED AT 10:51:17 ON

29 APR 2004
L1 1 S PHL1 VECTOR

FILE 'STNGUIDE' ENTERED AT 10:52:27 ON 29 APR 2004

FILE 'HCAPLUS' ENTERED AT 10:53:24 ON 29 APR 2004

FILE 'STNGUIDE' ENTERED AT 10:53:25 ON 29 APR 2004

FILE 'HCAPLUS, MEDLINE, BIOSIS, EMBASE, BIOTECHDS' ENTERED AT 10:53:35 ON 29 APR 2004

L2 1 S PHLB29

=> s alkaline lipase and dna

L3 22 ALKALINE LIPASE AND DNA

=> dup rem l3

PROCESSING COMPLETED FOR L3

L4 15 DUP REM L3 (7 DUPLICATES REMOVED)

=> focus l4

PROCESSING COMPLETED FOR L4

L5 15 FOCUS L4 1-

=> d l5 1-15 ibib ab

L5 ANSWER 1 OF 15 HCAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 2003:220114 HCAPLUS

DOCUMENT NUMBER: 138:349494

TITLE: Cloning and sequencing of genomic **DNA** encoding **alkaline lipase** from *Penicillium expansum* PF898

AUTHOR(S): Lin, Lin; Xie, Bi-feng; Yang, Guan-zhen; Shi, Qiao-qin; Lin, Qi-ying; Xie, Lian-hui; Wu, Song-gang; Wu, Xiang-fu

CORPORATE SOURCE: Institute of Virology, Fujian Agriculture and Forestry University, Fuzhou, 350002, Peop. Rep. China

SOURCE: Zhongguo Shengwu Huaxue Yu Fenzi Shengwu Xuebao (2003), 19(1), 12-16

CODEN: ZSHXF2; ISSN: 1007-7626

PUBLISHER: Zhongguo Shengwu Huaxue Yu Fenzi Shengwu Xuebao

DOCUMENT TYPE: Journal

LANGUAGE: Chinese

AB *Penicillium expansum* PF898 produces an alk. lipase (PEL) with industrial value. Based on the cloning of PEL cDNA by the methods of 3'RACE and 5'RACE, two primers were designed and the full-length genomic **DNA** was amplified from total **DNA** of the fungus by the method of PCR. The amplified **DNA** sequence of 1404 bp includess PEL coding area, 3' and 5' non-coding region. Anal. of the nucleotide sequence indicated that the genomic **DNA** of PEL (GenBank accession no. AF330635) was composed of 1135 bp and had six exons and five short introns (58 bp, 47 bp, 50 bp, 56 bp and 69 bp) The no. of introns of PEL is more than that of other fungi lipases that have been sequenced and they are all short introns. The 3' noncoding region is composed of 195 bp with an AATAAA sequence appeared at position 156 nt and the poly(A) tail is at the position 182 nt downstream of stop coden TGA. 5' non-coding region of 74 bp was sequenced and the TATA box was found at - 24 to - 27 nt of the gene. The homol. is about 39% .apprx. 49% between the genomic **DNA** sequence of PEL and that of other lipases from fungi. The homol. is about 42% .apprx. 57% between the introns of lipases from PEL and other several fungus.

L5 ANSWER 2 OF 15 HCAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 1998:692098 HCAPLUS

DOCUMENT NUMBER: 130:77772
 TITLE: Characterization of an **alkaline lipase** from *Proteus vulgaris* K80 and the DNA sequence of the encoding gene
 AUTHOR(S): Kim, Hyung-Kwoun; Oh, Tae-Kwang
 CORPORATE SOURCE: Applied Microbiology Research Group, KIST, Korea Research Institute of Bioscience and Biotechnology, Taejon, 305-600, S. Korea
 SOURCE: Proceedings of the World Conference on Oilseed and Edible Oils Processing, Istanbul, Oct. 6-10, 1996 (1998), Meeting Date 1996, Volume 2, 346-349.
 Editor(s): Koseoglu, S. S.; Rhee, K. C.; Wilson, Richard F. AOCS Press: Champaign, Ill.
 CODEN: 66VYAN
 DOCUMENT TYPE: Conference
 LANGUAGE: English
 AB Alk. lipase was isolated from a *Proteus vulgaris* K80 obtained from soil near a sewage treatment plant. Effects of temp. and pH. on the K80 lipase are reported. The gene was cloned and sequenced and homol. with related lipases is examd. The lid sequence and interfacial activation of K80 lipase are discussed.

L5 ANSWER 3 OF 15 HCAPLUS COPYRIGHT 2004 ACS on STN
 ACCESSION NUMBER: 1996:47727 HCAPLUS
 DOCUMENT NUMBER: 124:197009
 TITLE: Characterization of an **alkaline lipase** from *Proteus vulgaris* K80 and the DNA sequence of the encoding gene
 AUTHOR(S): Kim, Hyung-Kwoun; Lee, Jung-Kee; Kim, Hyoungman; Oh, Tae-Kwang
 CORPORATE SOURCE: Applied Microbiology Research Group, Korea Research Institute of Bioscience and Biotechnology, P.O. Box 115, Yusung, Taejon, 305-600, S. Korea
 SOURCE: FEMS Microbiology Letters (1996), 135(1), 117-21
 CODEN: FMLED7; ISSN: 0378-1097
 PUBLISHER: Elsevier
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB A facultatively anaerobic bacterium producing an extracellular alk. lipase was isolated from the soil collected near a sewage disposal plant in Korea and identified to be a strain of *Proteus vulgaris*. The mol. mass of the purified lipase K80 was estd. to be 31 kDa by SDS-PAGE. It was found to be an alk. enzyme having max. hydrolytic activity at pH 10, while fairly stable in a wide pH range from 5 to 11. The gene for lipase K80 was cloned in *Escherichia coli*. Sequence anal. showed an open reading frame of 861 bp coding for a polypeptide of 287 amino acid residues. The deduced amino acid sequence of the lipase gene had 46.3% identity to the lipase from *Pseudomonas fragi*.

L5 ANSWER 4 OF 15 HCAPLUS COPYRIGHT 2004 ACS on STN
 ACCESSION NUMBER: 1997:706004 HCAPLUS
 DOCUMENT NUMBER: 128:31829
 TITLE: **Alkaline lipase** of *Pseudomonas pseudoalcaligenes* and its encoding genes
 INVENTOR(S): Lien, Shun Fu; Chiu, Chien Ming; Chui, Kuan Hsiang
 PATENT ASSIGNEE(S): Daido Kufun Co., Ltd., Taiwan
 SOURCE: Jpn. Kokai Tokkyo Koho, 19 pp.
 CODEN: JKXXAF
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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JP 09275987 A2 19971028 JP 1996-119638 19960417
 PRIORITY APPLN. INFO.: JP 1996-119638 19960417
 AB A 2.9-kb DNA fragment contg. 2 open reading frames encoding alk. lipase is isolated from the chromosome of *Pseudomonas pseudoalcaligenes* strain F-111. Gene lipA and gene lipB encoding a 290- and a 340-amino-acid polypeptide, resp., are disclosed. Also claimed is the method for the prodn. of alk. lipase in transgenic *Escherichia coli*.

L5 ANSWER 5 OF 15 HCAPLUS COPYRIGHT 2004 ACS on STN
 ACCESSION NUMBER: 2003:397867 HCAPLUS
 DOCUMENT NUMBER: 139:64174
 TITLE: Cloning of an **alkaline lipase** gene from *Penicillium cyclopium* and its expression in *Escherichia coli*
 AUTHOR(S): Wu, Minchen; Qian, Zhikang; Jiang, Peihong; Min, Taishan; Sun, Chongrong; Huang, Weida
 CORPORATE SOURCE: Medical Department, Southern Yangtze University, Jiangsu, 214063, Peop. Rep. China
 SOURCE: Lipids (2003), 38(3), 191-199
 CODEN: LPDSAP; ISSN: 0024-4201
 PUBLISHER: AOCs Press
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB The gene encoding an alk. lipase of *Penicillium cyclopium* PG37 was cloned with four steps of PCR amplification based on different principles. The cloned gene was 1,480 nucleotides in length, consisted of 94 bp of promoter region, and had 6 exons and 5 short introns ranging from 50 to 70 nucleotides. The open reading frame encoded a protein of 285 amino acid residues consisting of a 27-AA signal peptide and a 258-AA mature peptide, with a conserved motif of Gly-X-Ser-X-Gly shared by all types of alk. lipases. However, this protein had a low homol. with lipases of *P. camembertii* (22.9%), *Humicola lanuginosa* (25.6%), and *Rhizomucor miehei* (22.3%) at the amino acid level. The mature peptide encoding cDNA was cloned and expressed in *Escherichia coli* on pET-30a for confirmation. A distinct band with a M.W. of 33 kDa was detected on SDS-PAGE. Results of a Western blot anal. and an enzyme activity assay verified the recombinant 33-kDa protein as an alk. lipase. Its catalytic properties were not changed when compared with its natural counterpart.
 REFERENCE COUNT: 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L5 ANSWER 6 OF 15 HCAPLUS COPYRIGHT 2004 ACS on STN
 ACCESSION NUMBER: 1998:414682 HCAPLUS
 DOCUMENT NUMBER: 129:64079
 TITLE: Cloning, expression and nucleotide sequence of an **alkaline lipase** gene from *Pseudomonas pseudoalcaligenes* F-111
 INVENTOR(S): Lin, Shuen-fuh; Chiou, Chien-ming; Chuang, Kuang-hsiang
 PATENT ASSIGNEE(S): Tatung Co., Ltd., Taiwan
 SOURCE: U.S., 22 pp.
 CODEN: USXXAM
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5766913	A	19980616	US 1996-606888	19960226
PRIORITY APPLN. INFO.:			US 1996-606888	19960226
AB Gene lipA from <i>P. pseudoalcaligenes</i> F-111 encoding alk. lipase is disclosed. This invention also discloses expressing lipA in <i>Escherichia coli</i> in order to obtain the lipase. The clone contg. the lipA gene was found to contain a further gene, lipB, which is proposed to control				

expression of lipA.

REFERENCE COUNT: 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L5 ANSWER 7 OF 15 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
ACCESSION NUMBER: 2004-05981 BIOTECHDS

TITLE: **Alkaline lipase** useful as laundry
detergent, isolated from *Vibrio metschnikovii* RH530 N-4-8;
recombinant enzyme production in *Escherichia coli*

AUTHOR: JIN G; JHON S; LEE H; RHO H

PATENT ASSIGNEE: CJ CORP

PATENT INFO: WO 2004001029 31 Dec 2003

APPLICATION INFO: WO 2003-KR1227 23 Jun 2003

PRIORITY INFO: KR 2002-35410 24 Jun 2002; KR 2002-35410 24 Jun 2002

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2004-082499 [08]

AB DERWENT ABSTRACT:

NOVELTY - An **alkaline lipase** (I) isolated from *Vibrio metschnikovii* RH530 N-4-8 comprising a fully defined sequence of 185 amino acids (S1) as given in the specification, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (1) polynucleotide (II) encoding (S1); (2) recombinant vector (III) comprising (II); (3) host cell (IV) transformed by (III); and (4) detergent comprising (I).

BIOTECHNOLOGY - Preparation: (I) is produced by culturing (IV) (claimed). Preferred Polynucleotide: (II) comprises a fully defined sequence of 555, 798 or 2578 base pairs as given in the specification. Preferred Recombinant Vector: (III) is pHL1, pHLB29 or pHAH38.

USE - (I) is useful as an enzyme for laundry detergent (claimed).

ADVANTAGE - (I) has high residual enzyme activity and high compatibility.

EXAMPLE - Culture medium comprising tryptone, yeast extract, sodium chloride in sodium carbonate buffer was used for culturing *Vibrio metschnikovii* RH530 N-4-8, at 30 degreesC. The cells were collected and treated with lysozyme to lyse the cell. The resultant product was treated with phenol and chloroform to remove protein, and a precipitate was removed by centrifugation. A *Vibrio* chromosomal DNA was obtained from the supernatant. The obtained chromosomal DNA was cut with a restriction enzyme HindIII to be recombined with a cloning vector pUC19, followed by transforming *Escherichia coli* HB101, thus cloning a DNA fragment containing a 3.2 kb **alkaline lipase** gene. The resulting recombinant vector was referred to as a vector pHL1. After treatment with the restriction enzyme HindIII, an electrophoresis with 1% agarose gel was performed. The agarose gel electrophoresis showed that the **alkaline lipase** gene was cloned. To confirm that a DNA fragment containing an **alkaline lipase** gene derived from *V. metschnikovii*, which is contained in a recombinant vector pHL1, is identical with the gene from *V. metschnikovii*, Southern blotting was performed. DNA of 3.2 kb was treated with an exonuclease Bal31 to subclone the same in a minimum length required for expression of a lipase. Production of the lipase was confirmed by formation of a clear halo, and the result of subcloning showed that 2.6 kb DNA fragment was necessary for lipase activity. The recombinant vector containing such a gene having a minimum length was referred to as pHLB29. DNA of 2.6 kb fragment was subcloned in a direction opposite to that of a SmaI site of pUC19, and referred to as pHAH38. Although the 2.6 kb DNA fragment was subcloned in a reverse direction relative to a lac promoter, pHAH38 produced a clear halo at a tricaprillin culture medium, confirming that an **alkaline lipase** promoter existed in the 2.6 kb DNA fragment and the promoter used when it is transcribed from *E. coli*. (35 pages)

L5 ANSWER 8 OF 15 MEDLINE on STN

ACCESSION NUMBER: 1998189963 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 9521811
 TITLE: **Alkaline lipase** from brain: is it the same enzyme as pancreatic lipase from pancreas?.
 AUTHOR: Tsujita T; Sumida M; Sumiyoshi M; Kameda K; Okuda H
 CORPORATE SOURCE: School of Medicine, Ehime University, Shigenobu, Ehime, Onsen-gun, 791-02, Japan.
 SOURCE: Archives of biochemistry and biophysics, (1998 Apr 1) 352 (1) 44-50.
 Journal code: 0372430. ISSN: 0003-9861.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199805
 ENTRY DATE: Entered STN: 19980514
 Last Updated on STN: 19980514
 Entered Medline: 19980504

AB A new **alkaline lipase** was detected in rat brain and its properties were compared with those of the well-characterized pancreatic lipase and pancreatic lipase-related protein 2. The activity of the **alkaline lipase** was determined using trioleoylglycerol emulsion at pH 8.0. Subcellular fractions were prepared from brain homogenates by differential centrifugation. Lipase activities of the cytosolic fraction (the supernatant obtained by differential centrifugation of 100,000g) were stimulated by addition of colipase and bile salts and inhibited by addition of an antibody against rat pancreatic lipase. The partially purified enzyme had an isoelectric point of pH 6.8, which was identical to that found for rat pancreatic lipase. The enzyme had interfacial activation and dependence on colipase in the presence of bile salts. The enzyme had no measurable phospholipase A activity. The band produced by the enzyme on SDS-polyacrylamide gel electrophoresis was identical to that of the rat pancreatic lipase when detected by immunoblotting analysis using an antibody against pancreatic lipase. These results show that pancreatic lipase such as **alkaline lipase** is in rat brain.
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L5 ANSWER 9 OF 15 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED.
 on STN

ACCESSION NUMBER: 1998220023 EMBASE
 TITLE: Development of a lipase fermentation process that uses a recombinant *Pseudomonas alcaligenes* strain.
 AUTHOR: Gerritse G.; Hommes R.W.J.; Quax W.J.
 CORPORATE SOURCE: W.J. Quax, Pharmaceutical Biology, University of Groningen, Antonius Deusinglaan 1, 9713 AV Groningen, Netherlands.
 w.j.quax@farm.rug.nl
 SOURCE: Applied and Environmental Microbiology, (1998) 64/7 (2644-2451).
 Refs: 39
 ISSN: 0099-2240 CODEN: AEMIDF
 COUNTRY: United States
 DOCUMENT TYPE: Journal; Article
 FILE SEGMENT: 004 Microbiology
 LANGUAGE: English
 SUMMARY LANGUAGE: English

AB *Pseudomonas alcaligenes* M-1 secretes an **alkaline lipase**, which has excellent characteristics for the removal of fatty stains under modern washing conditions. A fed-batch fermentation process based on the secretion of the **alkaline lipase** from *P. alcaligenes* was developed. Due to the inability of *P. alcaligenes* to grow on glucose, citric acid and soybean oil were applied as substrates in the batch phase and feed phase, respectively. The gene encoding the high-**alkaline lipase** from *P. alcaligenes* was isolated and characterized. Amplification of lipase gene copies in *P. alcaligenes* with

the aid of low- and high-copy-number plasmids resulted in an increase of lipase expression that was apparently colinear with the gene copy number. It was found that overexpression of the lipase helper gene, lipB, produced a stimulating effect in strains with high copy numbers (>20) of the lipase structural gene, lipA. In strains with lipa on a low-copy-number vector, the lipB gene did not show any effect, suggesting that LipB is required in a low ratio to LipA only. During scaling up of the fermentation process to 100 m3, severe losses in lipase productivity were observed. Simulations have identified an increased level of dissolved carbon dioxide as the most probable cause for the scale-up losses. A large-scale fermentation protocol with a reduced dissolved carbon dioxide concentration resulted in a substantial elimination of the scale-up loss.

L5 ANSWER 10 OF 15 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

ACCESSION NUMBER: 1998-01799 BIOTECHDS

TITLE: Lipolytic enzymes useful as detergent additives at high pH;
Gliocladium sp., Verticillium sp. or Trichophaea saccata
recombinant **alkaline lipase** expression
in Aspergillus oryzae

AUTHOR: Hirayama S; Taira R; Borch K; Sandal T; Halkier T; Oxenboll K
M; Nielsen B R

PATENT ASSIGNEE: Novo-Nordisk

LOCATION: Bagsvaerd, Denmark.

PATENT INFO: WO 9741212 6 Nov 1997

APPLICATION INFO: WO 1997-DK179 22 Apr 1997

PRIORITY INFO: DK 1996-501 25 Apr 1996; DK 1996-500 25 Apr 1996

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 1997-549717 [50]

AB A new lipolytic enzyme **DNA** fragment is cloned in a plasmid present in Escherichia coli DSM 10591, DSM 10590 or DSM 11298. The enzyme may be produced by: (1) growing Gliocladium sp. CBS 173.96, Verticillium sp. CBS 830.95, or Trichophaea saccata CBS 804.70; or (2) by isolating the **DNA**, combining the **DNA** with an expression signal in a vector and using this to transform a host (e.g. Aspergillus oryzae) which is cultured. The new **DNA** may be isolated by cloning a cDNA library from the strains in suitable vectors, transforming yeast cells and screening for expressed lipolytic activity in positive clones. The enzymes have specified sequences, or are at least 60% homologous. Enzymes have improved lipase (EC-3.1.1.3) and/or cutinase activity at alkaline pH, and improved oil hydrolysis activity on cotton/olive oil swatches. The enzymes may be used as: surfactant additives (e.g. non-dusting granulates or stabilized liquids) to remove fatty stains; for laundry and dishwashing at a high pH; or for interesterification, total hydrolysis of fats and oils, or optical isomer resolution. (71pp)

L5 ANSWER 11 OF 15 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

ACCESSION NUMBER: 1996-15062 BIOTECHDS

TITLE: **Alkaline lipase** for detergent use;
Botryosphaeria sp. or Guignardia sp. recombinant enzyme
production by vector expression in e.g. Aspergillus sp.,
for application as a surfactant additive

AUTHOR: Hirayama S; Halkier T

PATENT ASSIGNEE: Novo-Nordisk

LOCATION: Bagsvaerd, Denmark.

PATENT INFO: WO 9630502 3 Oct 1996

APPLICATION INFO: WO 1996-DK123 27 Mar 1996

PRIORITY INFO: DK 1995-830 14 Jul 1995; DK 1995-344 30 Mar 1995

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 1996-455350 [45]

AB An **alkaline lipase** (I, EC-3.1.1.3) which is immunologically reactive with an antibody raised against a purified lipase produced by Botryosphaeria sp. or Guignardia sp., and which has

optimum activity at pH 9-11 in the presence of 50 mM Ca²⁺ is claimed. Also claimed are: 1) a biologically pure culture of *Botryosphaeria* sp. CBS 102.95; and 2) a method for producing recombinant (I) by isolating an (I)-encoding **DNA** sequence from *Botryosphaeria* sp. or *Guignardia* sp., combining the sequence with expression elements in a vector, transforming a host (e.g. *Aspergillus* sp.) with the vector, culturing the transformant, and recovering (I) from the culture medium. (I) is produced by culturing (I)-producing *Botryosphaeria berengeriana* MAFF 06-45001, *Botryosphaeria berengeriana* F.sp. *pilicola* MAFF 06-45002, *Botryosphaeria dothidea* JCM 2733, 2735, 2736, 2737, *Botryosphaeria parva* ATCC 58191, *Botryosphaeria ribis* CBS 504.94 or ATCC 56125, *Botryosphaeria ribis* var. *chromogena* CBS 121.26, *Botryosphaeria xanthocephala* ATCC 60638, *Botryosphaeria* sp. CBS 102.95, *Guignardia laricina* IFO 7887 or 7888, or *Guignardia paulowniae* MAFF 03-05151. (43pp)

L5 ANSWER 12 OF 15 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
ACCESSION NUMBER: 1992-01713 BIOTECHDS

TITLE: New **alkaline lipase** from *Bacillus* species

Bacillus pumilus **DNA** sequence; vector expression
in transformant; potential application in surfactant
composition

PATENT ASSIGNEE: Kali-Chemie

PATENT INFO: DE 4111321 17 Oct 1991

APPLICATION INFO: DE 1991-111321 8 Apr 1991

PRIORITY INFO: DE 1990-12070 14 Apr 1990; DE 1991-111321 8 Apr 1991

DOCUMENT TYPE: Patent

LANGUAGE: German

OTHER SOURCE: WPI: 1991-311800 [43]

AB New lipases (EC-3.1.1.3), secreted by *Bacillus* sp., have a pH optimum in the alkaline range and a temp. optimum of 30-40 deg. Also new are: i. **DNA** sequences encoding the lipases having a protein sequence at least 70% (preferably 90%) homologous with a sequence (A); ii. transformation vectors containing the **DNA** sequences; iii. transformed microorganisms containing these vectors; and iv. *Bacillus pumilus* strains DSM 5776, DSM 5777 and DSM 5778. (A) contains 213 amino acids (including the signal peptide sequence) and is reproduced in the specification together with the encoding **DNA** sequence (793 bp). The lipase is useful in washing, cleaning, bleaching and dishwashing compositions, used at 30-40 deg. The compositions also preferably contain a protease. (29pp)

L5 ANSWER 13 OF 15 MEDLINE on STN

ACCESSION NUMBER: 1999155797 MEDLINE

DOCUMENT NUMBER: PubMed ID: 10036772

TITLE: Constituents of the tapetosomes and elaioplasts in *Brassica campestris* tapetum and their degradation and retention during microsporogenesis.

AUTHOR: Ting J T; Wu S S; Ratnayake C; Huang A H

CORPORATE SOURCE: Department of Botany and Plant Sciences, University of California, Riverside 92521, USA.

SOURCE: Plant journal : for cell and molecular biology, (1998 Dec) 16 (5) 541-51.

Journal code: 9207397. ISSN: 0960-7412.

PUB. COUNTRY: ENGLAND: United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

OTHER SOURCE: GENBANK-AF084554

ENTRY MONTH: 199903

ENTRY DATE: Entered STN: 19990402

Last Updated on STN: 20000303

Entered Medline: 19990325

AB In *Brassica* anthers during microsporogenesis, the tapetum cells contain two abundant lipid-rich organelles, the tapetosomes possessing oleosins

and triacylglycerols (TAGs), and the elaioplasts having unique polypeptides and neutral esters. *B. campestris*, for its simplicity of possessing only the AA genome and one predominant oleosin of 45 kDa, was studied. In the developing anthers, the lipids and proteins of the tapetosomes and elaioplasts were concomitantly accumulated but selectively degraded or retained. Upon incubation of isolated tapetosomes in a pH-5 medium, the predominant 45 kDa oleosin underwent selective enzymatic proteolysis to a 37 kDa fragment, which was not further hydrolyzed upon prolonged incubation. The unreacted 45 kDa oleosin was retained in the organelles, whereas the 37 kDa fragment was released to the exterior. The fragment would become the predominant 37 kDa polypeptide in the pollen coat. Isolated tapetosomes did not undergo hydrolysis of the TAGs upon incubation in media of diverse pHs. An **alkaline lipase** in the soluble fraction of the anther extract was presumed to be the enzyme that would hydrolyze the tapetosome TAGs, which disappeared in the anthers during development. The tapetum elaioplasts contained several unique polypeptides of 31-36 kDa. The gene encoding a 32 kDa polypeptide was cloned, and its deduced amino acid sequence was homologous to those of two proteins known to be present on the surface of fibrils in chromoplasts. Upon incubation of isolated elaioplasts in media of diverse pHs, the organelle polypeptides were degraded completely and most rapidly at pH 5, whereas the neutral esters remained unchanged; these neutral esters would become the major lipid components of the pollen coat. The findings show that the constituents of the two major tapetum organelles underwent very different paths of degradation, or modification, and transfer to the pollen surface.

L5 ANSWER 14 OF 15 MEDLINE on STN
 ACCESSION NUMBER: 2001555126 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 11601622
 TITLE: Characterization of the lipA gene encoding the major lipase from *Pseudomonas aeruginosa* strain IGB83.
 AUTHOR: Martinez A; Soberon-Chavez G
 CORPORATE SOURCE: Departamento de Microbiologia Molecular, Instituto de Biotecnologia, Universidad Nacional Autonoma de Mexico, Cuernavaca, Morelos.
 SOURCE: Applied microbiology and biotechnology, (2001 Sep) 56 (5-6) 731-5.
 Journal code: 8406612. ISSN: 0175-7598.
 PUB. COUNTRY: Germany: Germany, Federal Republic of
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 OTHER SOURCE: GENBANK-AF237723
 ENTRY MONTH: 200204
 ENTRY DATE: Entered STN: 20011017
 Last Updated on STN: 20020419
 Entered Medline: 20020418

AB The lipases produced by *Pseudomonas* have a wide range of potential biotechnological applications. *Pseudomonas aeruginosa* IGB83 was isolated as a highly lipolytic strain which produced a thermotolerant and **alkaline lipase**. In the present work, we have characterized the *P. aeruginosa* IGB83 gene (lipA) encoding this enzyme. We describe the construction of a lipA mutant and report on the effect of two carbon sources on lipase expression.

L5 ANSWER 15 OF 15 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
 ACCESSION NUMBER: 2001-06770 BIOTECHDS
 TITLE: Over-expression and properties of a purified recombinant *Bacillus licheniformis* lipase: a comparative report on *Bacillus* lipases;
 enzyme expression in *Escherichia coli*, purification and characterization
 AUTHOR: Nthangeni M B; Patterton H G; van Tonder A; Vergeer W P; Litthauer D

CORPORATE SOURCE: Univ.Orange-Free-State
LOCATION: Department of Microbiology and Biochemistry, University of
the Free State, P.O. Box 339, Bloemfontein, 9300, South
Africa.
Email: nthangen@micro.nw.uovs.ac.za
SOURCE: Enzyme Microb.Technol.; (2001) 28, 7-8, 705-12
CODEN: EMTED2
ISSN: 0141-0229
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Polymerase chain reaction using degenerate primers was used to clone the
gene encoding extracellular lipase (EC-3.1.1.3) from Bacillus
licheniformis strain DSM 12369 or strain UOFS. Genomic DNA
encoding the mature lipase was subcloned into expression vector plasmid
pET20b(+). High level expression of the lipase by Escherichia coli
JM109(DE3) cells harboring the vector was observed upon induction with
IPTG at 30 deg. The recombinant lipase was expressed as a fusion protein
with a C-terminal hexahistidine affinity tail. Single-step purification
was achieved by metal chelate affinity chromatography on Ni-2+-chelated
nitriloacetic acid. The purified enzyme had a specific activity of 130
U/mg with p-nitrophenyl palmitate as substrate. Optimum activity was
shown at pH 10-11.5, with high stability at alkaline pH values up to 12.
The enzyme was active toward p-nitrophenyl esters of short to long chain
fatty acids, with a marked preference for esters with C6 and C8 acyl
groups. Amino acid sequence similarity was shown to lipases from
Bacillus subtilis and Bacillus pumilus. It is proposed that Bacillus
lipases be classified into 2 distinct subfamilies of their own. (29 ref)

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FILE 'STNGUIDE' ENTERED AT 10:50:38 ON 29 APR 2004

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29 APR 2004

L1 1 S PHL1 VECTOR

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FILE 'STNGUIDE' ENTERED AT 10:53:25 ON 29 APR 2004

FILE 'HCAPLUS, MEDLINE, BIOSIS, EMBASE, BIOTECHDS' ENTERED AT 10:53:35 ON
29 APR 2004

L2 1 S PHLB29

L3 22 S ALKALINE LIPASE AND DNA

L4 15 DUP REM L3 (7 DUPLICATES REMOVED)

L5 15 FOCUS L4 1-

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